REMARKS

Rejection Under 35 USC 112, First Paragraph

Claims 1-14, 16-34, 36-54, 56-71, 73-75 and 81 have been rejected under 35 USC 112, first paragraph. More specifically, the Patent Office states:

> the specification .... does not reasonably provide enablement for heat shock/ubiquitin fusion proteins fused to "all" T or B cell epitopes, self epitopes, structural mimics of biomolecules, microbial epitopes, or heat shock/ubiquitin fusion proteins which are modified at the C-terminus to inhibit cleavage by ubiquitin-specific protease or which is postranslationally modified by the addition of fatty acids, or a ubiquitin fusion protein linking two domains of secondary structure ...

In response to this rejection, Claims 1, 20, 41, and 58 have been canceled and their subject matter incorporated into now independent Claims 2, 21 and 59 by amendment, limiting the claims of the invention to ubiquitin fusion proteins. Claims 2, 21 and 59 have further been amended to specifically recite that the claimed ubiquitin fusion protein is characterized by the ability to stimulate an immune response to heterologous epitope(s) contained therein. The Exemplification provides evidence that a ubiquitin fusion protein of the present invention is immunogenic for at least one of the epitopes contained therein. This has been demonstrated for three independent epitopes, V3 of HIV gp120, GnRH epitopes, and growth hormone epitopes. Example 1 demonstrates that the HIV epitope V3 of the gp120 molecule is highly antigenic when in the form of a ubiquitin fusion protein of the present invention (see page 33, line 1-20, Table 1, and page 36, line 6-14). Inclusion of a T-cell epitope in the fusion protein was shown to further enhance the antigenicity of the V3 epitope (Table 1). Example 3 demonstrates that epitopes from GnRH are also highly immunogenic in the form of a ubiquitin fusion protein of the present invention. Example 6 and 7 demonstrate that self epitopes (derived from GnRH or growth

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hormone) are immunogenic when incorporated into a ubiquitin fusion protein.

The selection of appropriate epitopes for use in the ubiquitin fusion protein of Claims 2, 21, 42, and 59 is within the ability of one of average skill in the art. Applicants' disclosure provides methods for verifying that the produced ubiquitin fusion protein exhibits the specified characteristics of being immunogenic for an epitope(s) contained therein. The fusion of ubiquitin to an epitope containing segment which has two or more copies of any known identical epitopes is within the ability of one of average skill in the art through routine procedures of genetic engineering. The specific use of ubiquitin as the heat shock protein and any known T cell or B cell epitope, self epitope, structural mimic of a biomolecule, or microbial epitope to produce a fusion protein of the present invention is also within the ability of one of average skill in the art through no more than routine experimentation. An abundance of procedures for genetic engineering of nucleic acids to produce sequences which encode fusion proteins are regarded as standard in the art. Any sequences which are required are readily available or easily determined from the art. An abundance of procedures for manipulating nucleic acid sequences *in vitro* to produce translated protein products are also available and used as standard practice in the art. Methods for modifying the C-terminus of a ubiquitin molecule (as a portion of an ubiquitin fusion protein) to inhibit cleavage of fusions located on the C-terminus of the ubiquitin molecule is known in the art as well as explained in the present disclosure. Methods for post translational addition of fatty acids to a protein (e.g. by incorporating specific modification signal sequences and subjecting proteins to known chemical reactions) are well known in the art, and the ubiquitin fusion protein of the present invention has no known properties which prevent the application of these methods.

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For clarity, Claims 16, 36, and 56 have been amended to specifically recite that the ubiquitin fusion protein of the present invention is generated from insertion of epitope containing segments at internal fusions sites of ubiquitin which link two ubiquitin domains of secondary structure, β-strand and α-helix. Identification of such sites is also within the ability of one of skill in the art given the high level of understanding in the art of the structure of the ubiquitin protein.

With respect to Claims 11-12, 31-32, 51-52, and 68-59, the Patent Office states:

> The breadth of these claims are way beyond what is disclosed in the specification. No where in the specification is there any disclosure of structural mimics of biomolecules or microbial epitopes... absent any guidance from the specification of the structural mimics of biomolecules and microbial epitopes, the skilled artisan could not predict which structural mimics of biomolecules or microbial epitopes would be reasonably expected to have the desired biological activities.

Applicants respectfully disagree with the statements of the above quoted passage as they apply to the present invention. It is respectfully submitted that the "desired biologic activity of an epitope" regarding the present invention is whether the epitope is antigenic in the context of a ubiquitin fusion protein. Applicants' disclosure provides methods for making such a determination, and those methods are considered routine and within the ability of one of average skill in the art.

Regarding Claims 74 and 75, the Patent Office states:

> ... it would require undue experimentation to determine which nucleic acids encoding fusion proteins having the desired biological activity, would be encompassed by the scope of the claims.

This rejection is respectfully traversed. Applicants' arguments above regarding the enablement of claims directed towards ubiquitin fusion protein apply equally to the rejection of Claims 74 and 75. As discussed above, Applicants maintain

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that the production of a ubiquitin fusion protein recited by amended Claims 2, 21, 42, and 59 is within the ability of one of skill in the art. The determination of nucleic acid sequences which encode a specific fusion protein so identified is accomplished by routine procedures which are standard in the art.

It is respectfully pointed out that, to the best of Applicants' understanding, neither Claims 74 nor Claim 75 qualifies as a "single means" claim, as defined in In re Hyatt. More specifically, Judge Rich defines a single means claim as "a claim drafted in "means-plus-function" format yet reciting only a single element instead of a combination." Claim 74 specifies a DNA construct encoding a ubiquitin fusion protein as described in Claim 21, 42, or 59, and Claim 75 specifies a cell containing said DNA construct.

Rejection Under 35 USC 112, Second Paragraph

Claims 1-6, 8, 15, 16, 20, 22-25, 27-35, 37, 38, 40, 41, 43-46, 48, 50-55, 57, 58, 60-63, 65-68, and 71-73 have been rejected under 35 USC 112, second paragraph. More specifically, the Patent Office states:

> claims 1, 3, 8, 20,22, 27-38, 40, 41, 43, 48, 50-55, 57, 58, 60, 65-68, and 71-73, are vague and indefinite for the recitation of "epitope-containing segment...", the meaning of the phrase is unclear the metes and bounds of these epitope containing segments are not ascertainable. The issue here is how large should these segments be.

The above described amendment of Claims of the present invention in response to the 35 USC 112, first paragraph rejection, to specifically recite that the ubiquitin fusion protein is "immunogenic for the heterologous epitope contained therein" functionally limits the metes and bounds of inserted epitope-containing segments. Such functional limitations of the epitope-containing segments are described in the specification as preserving systemic tolerance and functional tolerance to the ubiquitin fusion protein (page 7, line 12 - 34).

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The Patent Office further states:

> With respect to Claims 4-6, 23-25, 44-46, and 61-63 ... these claims are vague and unclear.

In response to this rejection, Claims 5, 6, 24, 25, 45, 46, 62, and 63 have been amended to specifically refer to the C-terminal ubiquitin subdomain as an "additional C-terminal subdomain".

Regarding the remaining claims rejected under 35 USC 112, second paragraph, the Patent Office further states:

> With respect to claims 20 which recite "two or more non-contiguous epitopes", it is unclear whether the non-contiguous epitopes are attached to different sites of the ubiquitin protein or whether one is fused to the ubiquitin protein and others are fused to the first epitope.... Claim 15 is indefinite because the "plurality of identical epitopes.." lacks antecedent basis. Claims 16, 36, and 56 are indefinite because the "internal fusion sites..."lacks antecedent basis. Claims 81 and 83 are vague and indefinite. The phrase "Ubiquitin having the peptide fused via its N-terminus to the C-terminal residue of ubiquitin..." is confusing because it is unclear if "its" relates to ubiquitin or to the peptide.... It is suggested that (claims 81 and 83) be amended to recite "consisting" which is closed language or "comprising" which is open language.

Regarding the rejection of Claim 20, Applicants respectfully point out that Claim 20 does not recite "two or more non-contiguous epitopes", but rather recites "two or more non-contiguous epitope-containing segments". The description of the segments as 'non-contiguous', is generally accepted as meaning not touching or connected in an unbroken sequence. Applicants regard this description as clearly indicating that the epitope-containing segments are located at different sites of the ubiquitin protein, rather than falling one after another at the same site in the ubiquitin protein. Claim 15 has been amended to remove the objectionable term 'plurality'. Claims 16, 36, and 56 have been amended to appropriately introduce the term 'internal fusion site'. For clarity, Claims 81 and 83 have been amended to no longer contain term 'ubiquitin having'.

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Rejection Under 35 USC 102(b) and (e)

Claims 1, 41, and 58 have been rejected under 35 USC 102(b) as being anticipated by Lussow et al. More specifically the Patent Office states:

> Lussow et al. teach a heat shock protein (hspR65) coupled to a synthetic immunogenic peptide consisting of 40 Asn-Ala-Asn-Pro (NANP) repeats from the repetitive region of the major antigen, the circumsporozite (CS) protein that covers the external surface of the human malaria parasite, (Page 2297, column 2, lines 19028). Claims 1, 41, and 58 of the present application recite "... a heat shock protein fused to epitopes ..." Therefore, the heat shock protein fused to the synthetic immunogenic peptide taught by Lussow et al reference anticipates claims 1, 41, and 58.

This rejection has been obviated by the above described amendment which limits Applicants' claims to ubiquitin fusion proteins. Furthermore, the ability of a heat shock protein such as ubiquitin to serve as a scaffold for presenting epitope fused by the means of the present invention would not have been obvious to one of skill in the art from the disclosure of Lussow et al., due to the difference in fusion construction and especially since Lussow et al. indicated that smaller sized heat shock proteins did not function as immunogenic carrier proteins (page 2300, column 1, second paragraph). Lussow et al. teach a heat shock protein covalently coupled via conjugation by exposure to glutaraldehyde, which crosslinks the NH_2 side chains of the carrier and antigen via a Schiff base. This is not a true fusion protein as described in the present disclosure, but rather is a protein conjugate as defined in the art. The fusion protein of the present invention is a fusion of a heat shock protein to epitopes by an single contiguous amide linkage (page 7, line 17-19) as the term is widely used in the art.

Claims 2, 3, 8, 14, 42, 43, 54, 59, and 60 have been rejected under 35 USC 102(e) as being anticipated by Vierstra et al. More specifically, the Patent Office states:

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> Vierstra et al teach ubiquitin fusion proteins where target proteins are attached to the carboxyl terminal of a ubiquitin molecule, such as the conjugation of ubiquitin to immunoglobulins ... therefore, the ubiquitin fusion proteins taught by Vierstra et al reference anticipate claims 2-3, 8, 14, 42, 43, 54, 59 and 60.

This rejection is respectfully traversed. Vierstra et al. teach an E2 (ubiquitin conjugating enzyme) fusion protein, not an ubiquitin fusion protein. Ubiquitin proteins discussed in the disclosure of Vierstra et al. are either 1) attached to heterologous proteins via lysine chains of the heterologous protein, not a single, contiguous amide linkage fusion protein of the present invention; 2) conjugated to immunoglobulins (column 11, line 49 to column 12, line 3), to produce a protein conjugate rather than a true fusion protein; or 3) attached to E2 through a thiol ester linkage to a cysteine within E2 (column 4, line 1-4) which is also not a true fusion protein as described in the present disclosure (as discussed above).

Claims 10, 13, 28, 33, 37, 50, 53, 67, and 70 have been rejected under 35 USC 102(b) as being anticipated by Mouritsen et al. More specifically the Patent Office states:

> Mouritsen et al. teach the attachment of one or more foreign T cell epitopes into the highly conserved self protein ubiquitin. ... the injection of these ubiquitin fusion proteins into mice elicited strong antibody response against the ubiquitin self protein. Claims 10, 13, 28, 33, 37, 50, 53, 57, 65, 67, and 70 recite " .... ubiquitin fusion protein attached to T cell epitopes and self epitopes". Thus ubiquitin fusion proteins with T cell epitopes taught by Mouritsen et al anticipate claims 10, 13, 28, 33, 37, 50, 53, 57, 65, 67, and 70.

This rejection is respectfully traversed. Mouritsen et al. teach a fusion protein resulting from the insertion of a single copy of a T cell epitope internally into a single ubiquitin amino acid sequence to produce a fusion protein which is antigenic for the ubiquitin portion. Applicants' claims specify a ubiquitin fusion protein which results from fusion of multiple epitopes to

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a single heat shock protein. Claims which specify a ubiquitin fusion protein which results from fusion of one or more epitopes to the heat shock protein, specify that fusion occurs on the N-terminus of ubiquitin. Therefore, Applicants' claims are not anticipated by Mouritsen et al. Furthermore, the ubiquitin fusion protein of the present invention is not obvious in light of the fusion protein of Mouritsen et al. because Applicants' fusion protein is antigenic for the heterologous epitopes, whereas the fusion protein of Mouritsen et al. is antigenic for ubiquitin. The functional 'self-epitopes' of Mouritsen et al. were epitopes present on the ubiquitin molecule, not epitopes which were fused to the ubiquitin molecule. Epitopes fused to the ubiquitin molecule in Mouritsen et al. were necessarily foreign. This also differs from the fusion protein of the present invention.

Rejection Under 35 USC 103(a)

Claims 15, 35, 55, 72, 81 and 83 have been rejected under 35 USC 103(a) as unpatentable over Van der zee et al. in view of Vannier et al. More specifically the Patent Office states:

> it would have been obvious to one of ordinary skill in the art to modify the GnRH fusion protein taught by Van der zee et al by fusing GnRH to ubiquitin as taught by Vannier et al because ubiquitin is a small highly conserved protein that is found in all eukaryotic cells and that does not induce immune response in animals.

This rejection is respectfully traversed. Vannier et al. use a ubiquitin-human FSHR fusion protein to generate monoclonal antibodies in mice to FSHR. The ubiquitin fusion protein of the present invention would not have been obvious to one of skill in the art upon reading Vannier et al. due to the lack of information provided regarding the construction of the ubiquitin fusion protein used by Vannier. Vannier et al. make no reference as to the construction of the fusion protein or why it was used (e.g. was a ubiquitin-FSHR fusion protein which promotes cleavage of the ubiquitin moiety upon inoculation used simply to increase

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the yield of FSHR produced in their prokaryotic system?) Vannier et al. refers to Loosfelt et al. *(Proc. Natl. Acad. Sci. 89*: 3765-3769 (1992)) in the description of the ubiquitin fusion protein. Loosfelt et al. describes the use of a β-galactosidase fusion protein for use as an immunogen, and describes the use of a ubiquitin fusion protein to screen for monoclonal antibodies, not as an immunogen. Moreover, Loosfelt et al. fails to provide any information regarding the construction of the ubiquitin fusion protein.

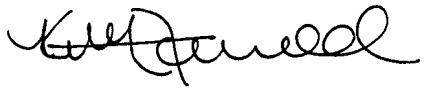
Objection to Claims Under 37 CFR 1.821 (d)

In order to comply with 37 CFR 1.821 (d), the recitation of amino acid sequences have been removed from Claim 81 and 83 by amendment.

Summary

In light of the above amendment and remarks, reconsideration of the subject patent application is respectfully requested.

Respectfully submitted,



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